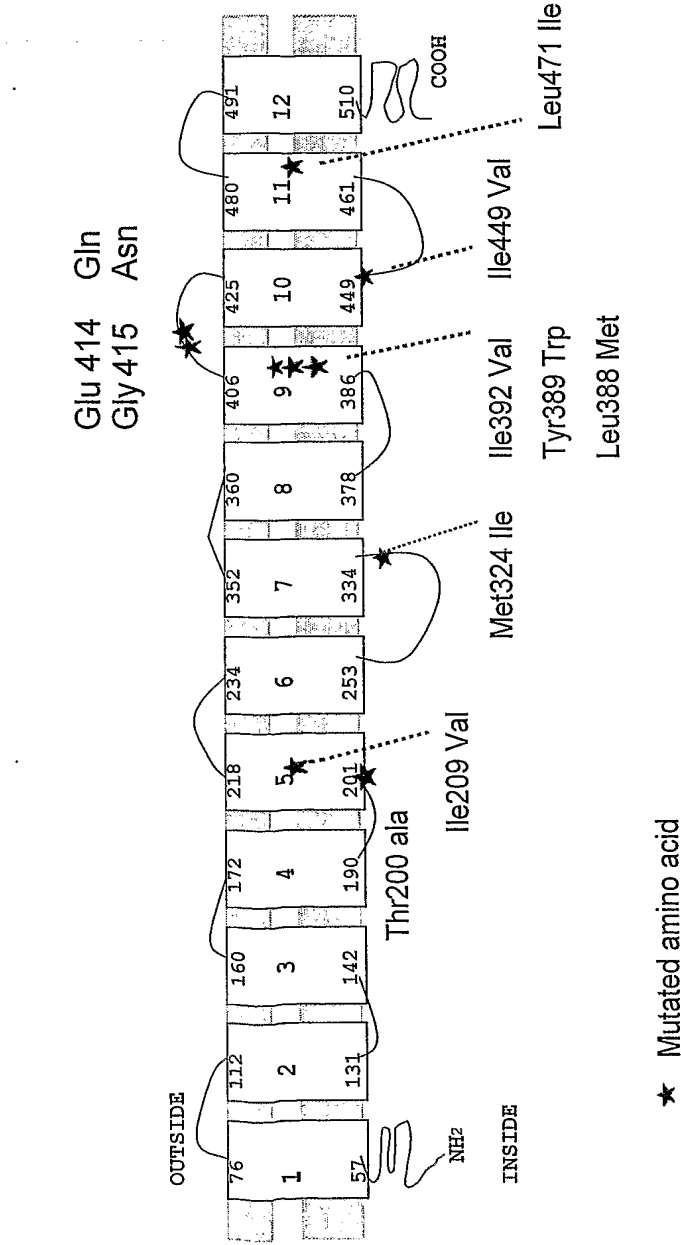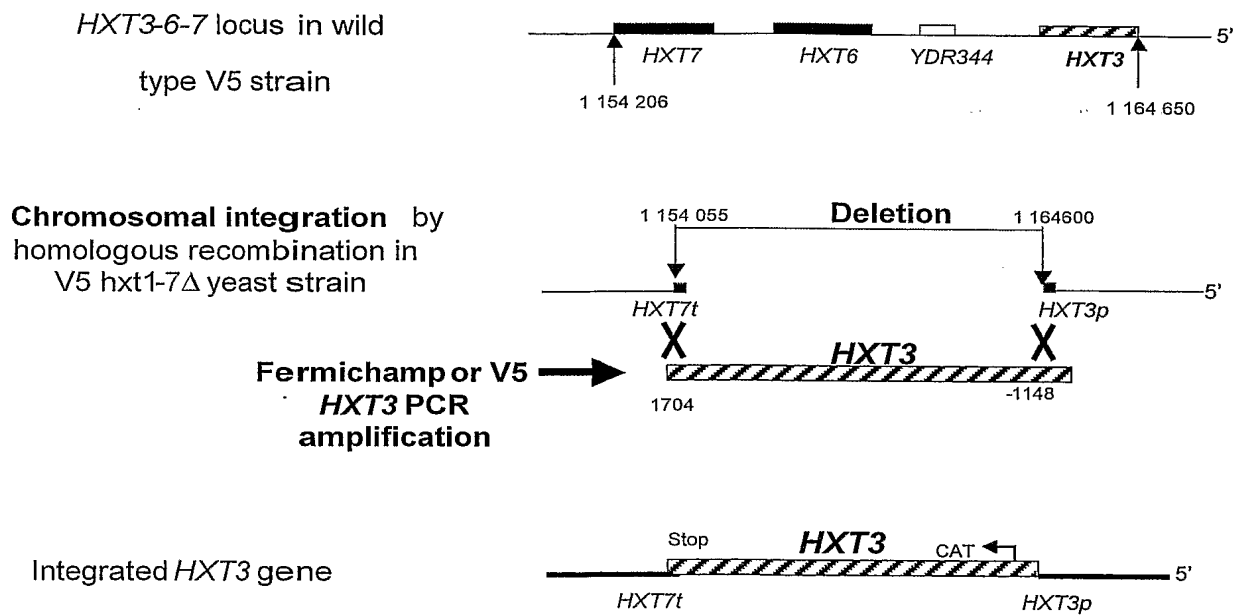Figure 1: Localisation of mutations in Fermichamp HXT3

Figure 2A Construction of V5 strains with integrated *HXT3* genes

HXT3 integration in V5 *hxt1-7Δ* strain

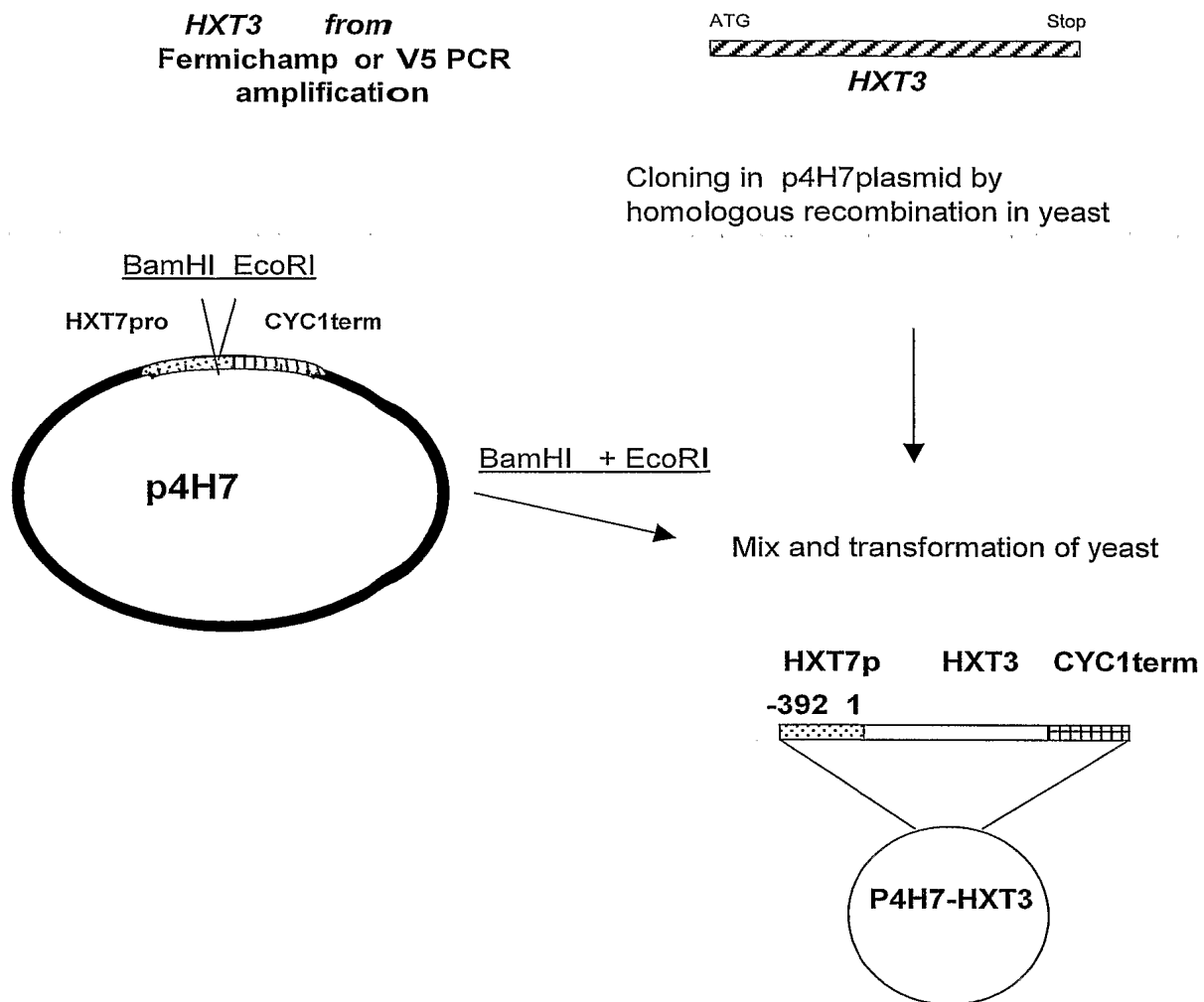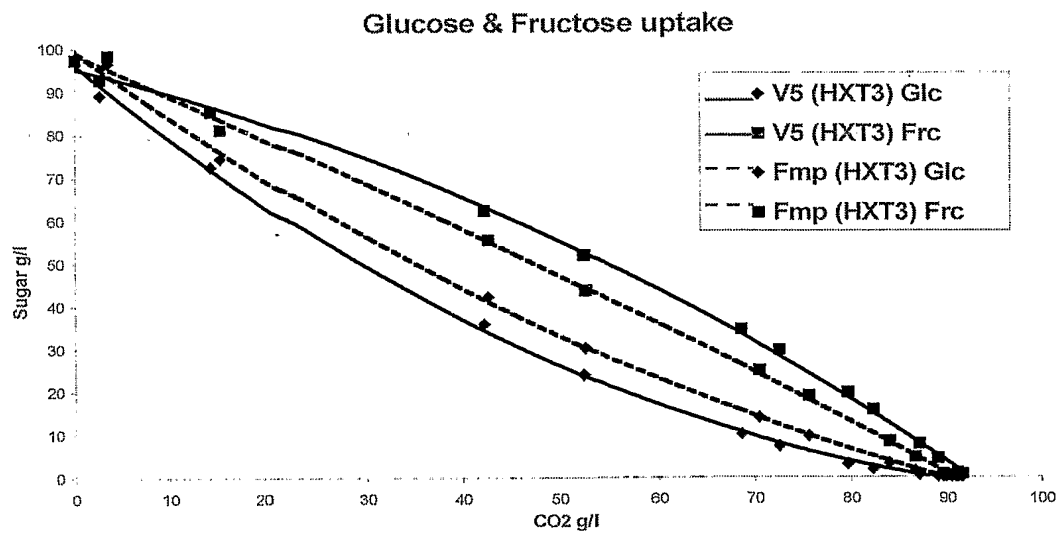Figure 2B : *HXT3* ORF cloning in multicopy plasmid p4H7

**HXT3   from
Fermichamp or V5 PCR
amplification**

ATG   Stop

HXT3

Cloning in p4H7 plasmid by
homologous recombination in yeast

<u>BamHI EcoRI</u>

HXT7pro   CYC1term

p4H7

<u>BamHI + EcoRI</u>

Mix and transformation of yeast

HXT7p   HXT3   CYC1term

-392 1

P4H7-HXT3

Figure 3A glucose and fructose utilisation by *HXT3* (**V5** or **Fmp)** single copy
gene expression

Glucose & Fructose uptake

Figure 3B : glucose/fructose ratio of *HXT3* (**V5** or **Fmp**) single copy gene expression
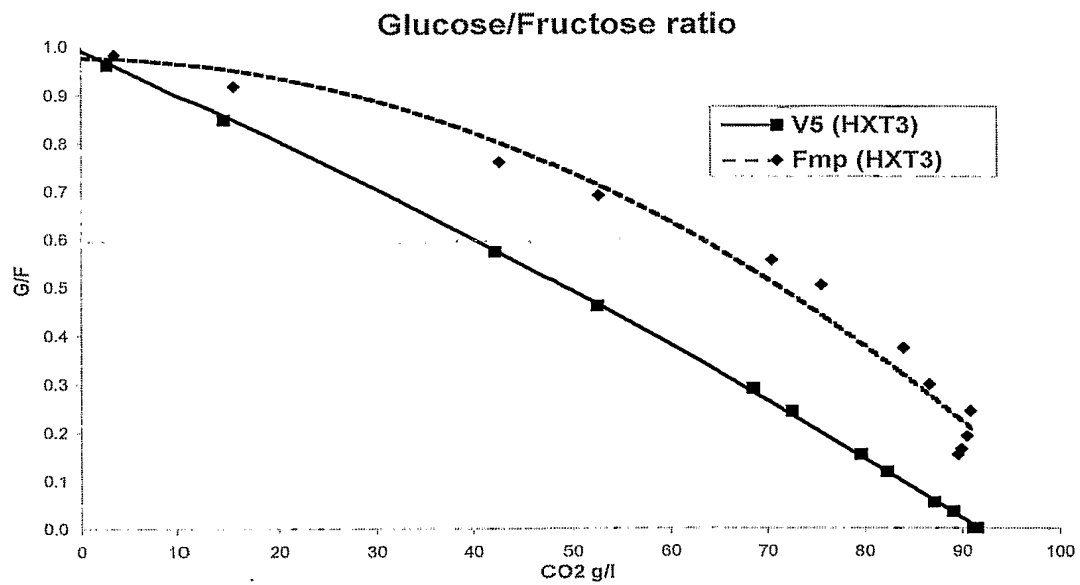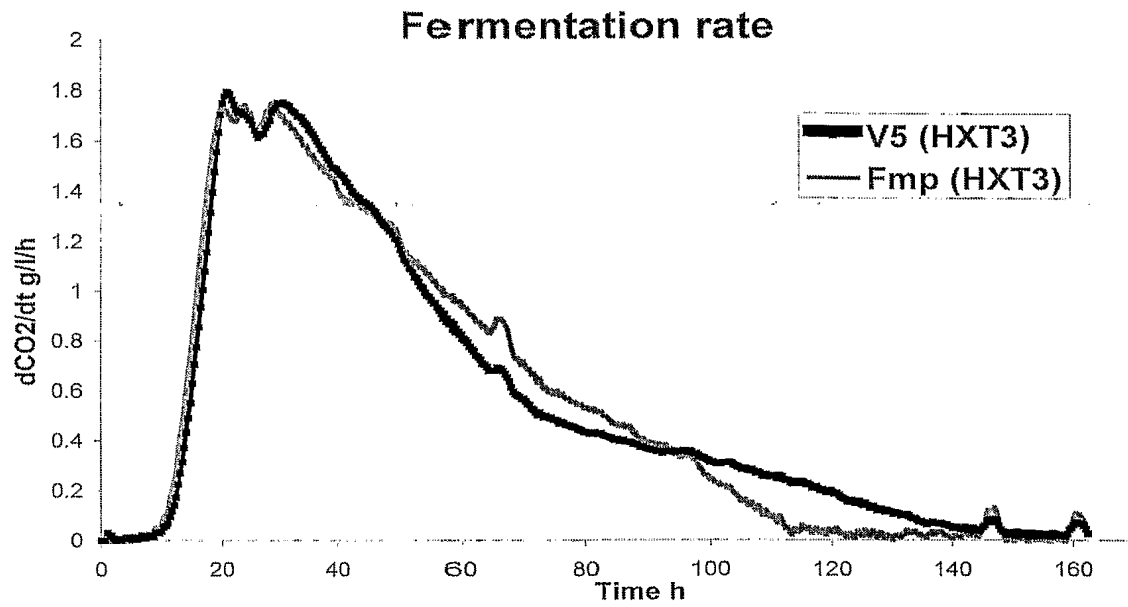
Glucose/Fructose ratio

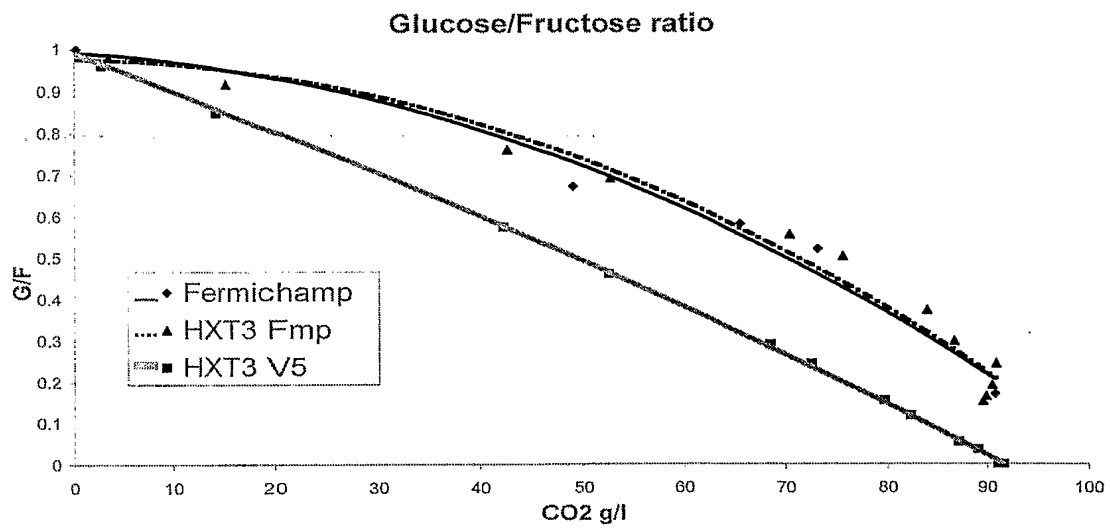Figure 3C : fermentation rate of *HXT3* (**V5** or **Fmp**) single copy gene expression

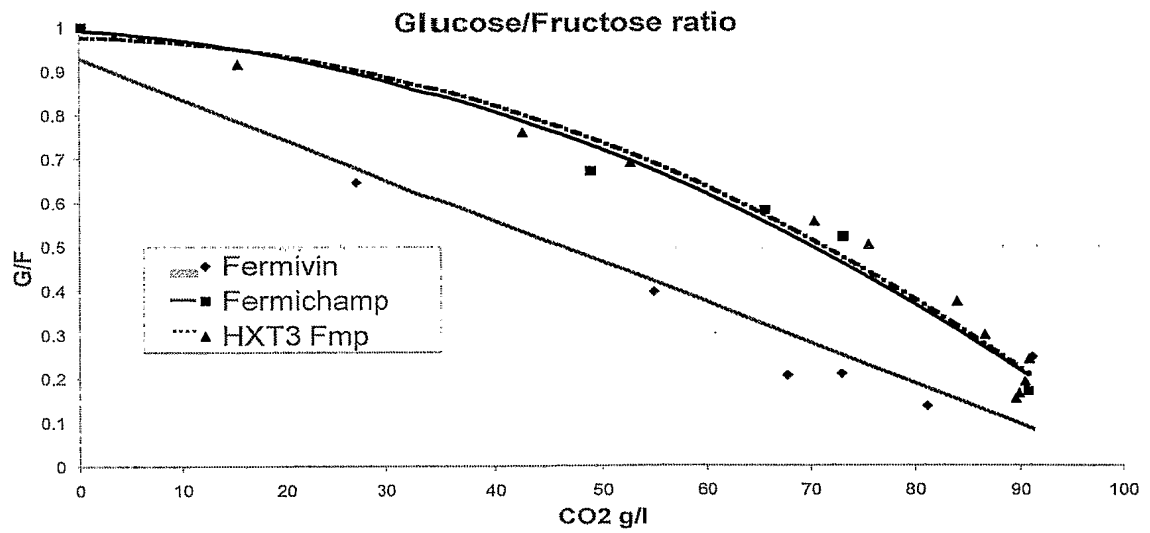Figure 3C : fermentation rate of *HXT3* (V5 or Fmp) single copy gene expression

<u>Figure 3D</u>

Comparison of Glucose / Fructose ratio between Fermichamp & *HXT3* (**V5** or

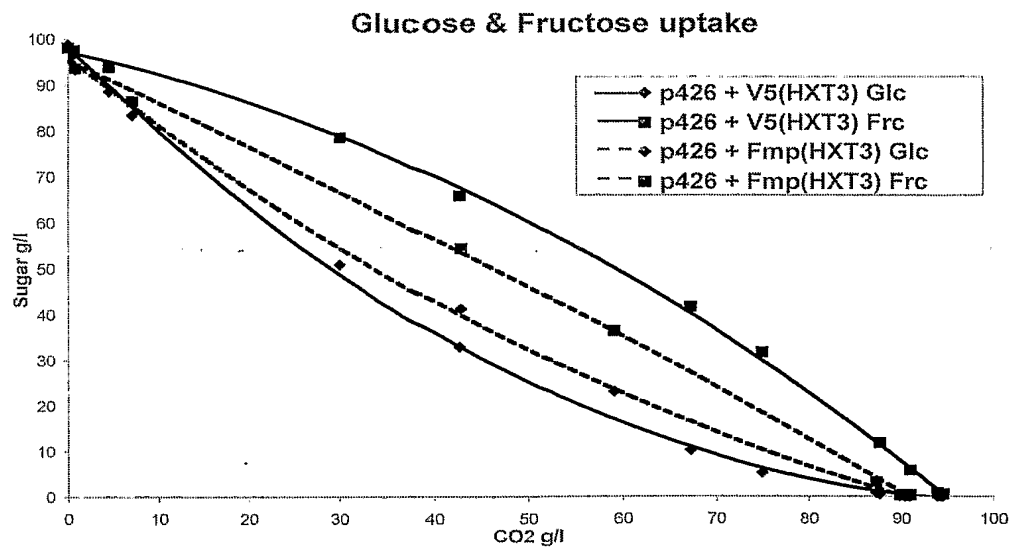**Fmp**) single copy gene expression

Glucose/Fructose ratio

Figure 3 E : Comparison of Glucose / Fructose ratio between Fermichamp,
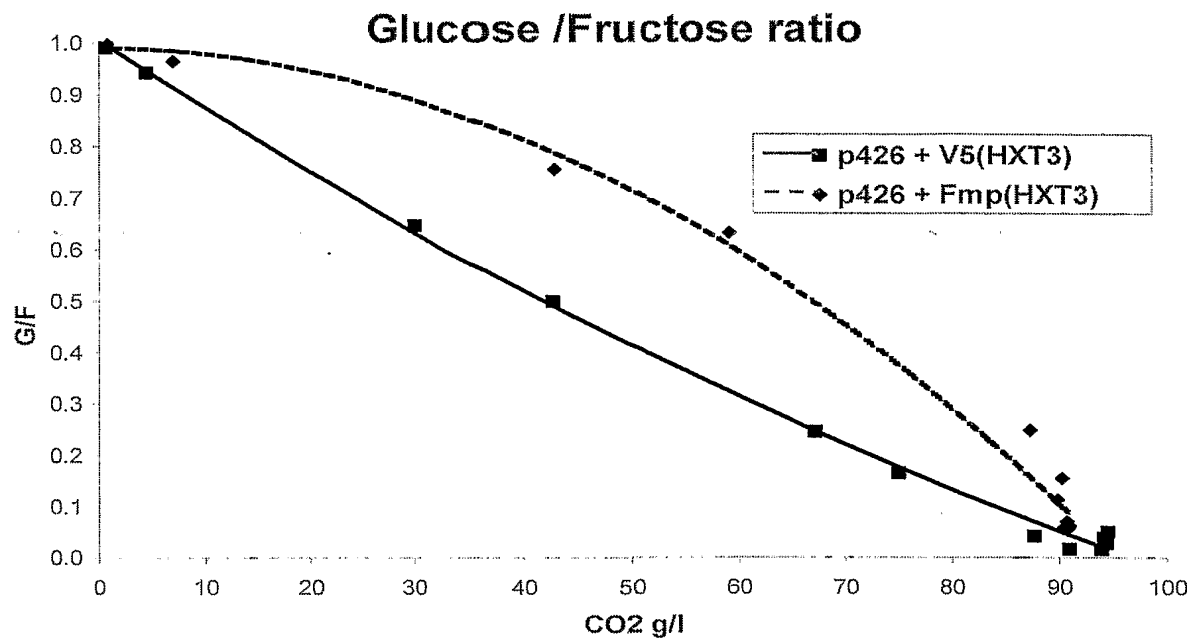Fermivin & *HXT3* **Fmp** single copy gene expression

Figure 4A : glucose and fructose utilisation by multicopy overexpression of *HXT3* (V5 or Fmp)

Glucose & Fructose uptake

Legend: p426 + V5(HXT3) Glc; p426 + V5(HXT3) Frc; p426 + Fmp(HXT3) Glc; p426 + Fmp(HXT3) Frc

Figure 4B : glucose/fructose ratio by multicopy overexpression of *HXT3* (V5 or Fmp)

Glucose /Fructose ratio
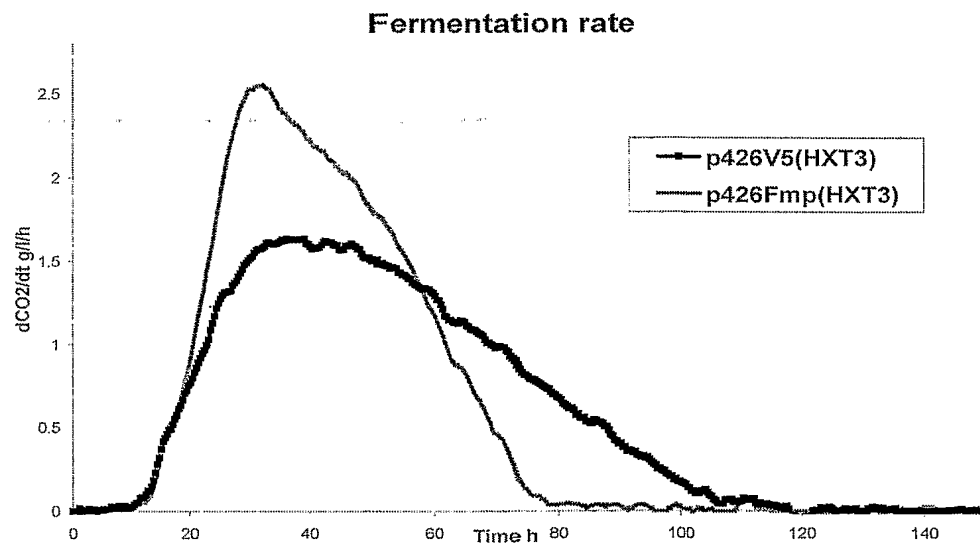
Figure 5 : Multicopy overexpression of *HXT3* (V5 or Fmp) on Glucose + Fructose (50/50) must (200g/l)
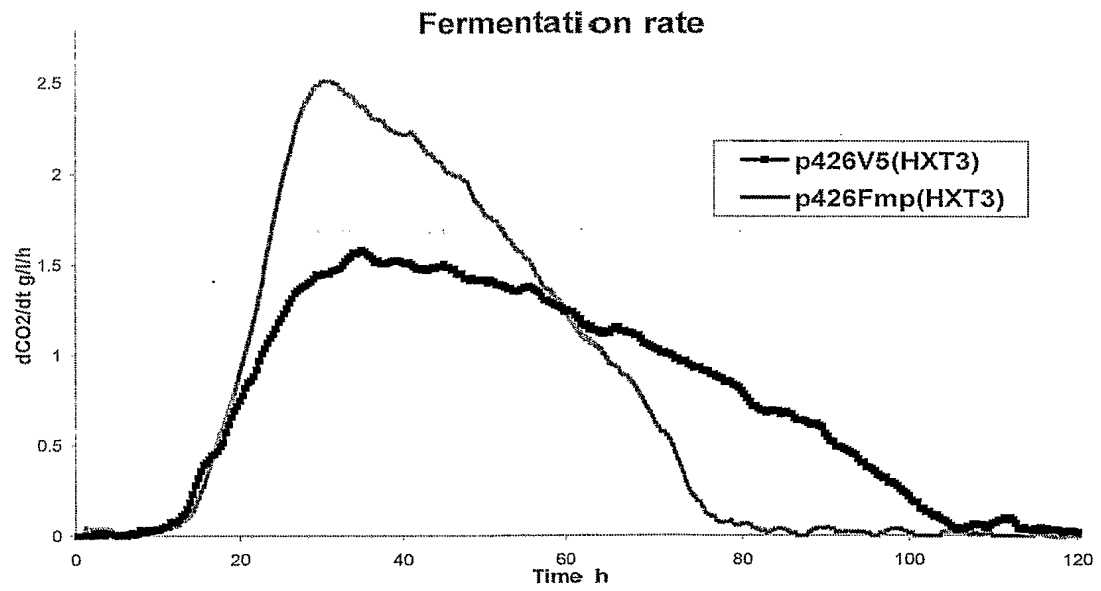
Fermentation rate
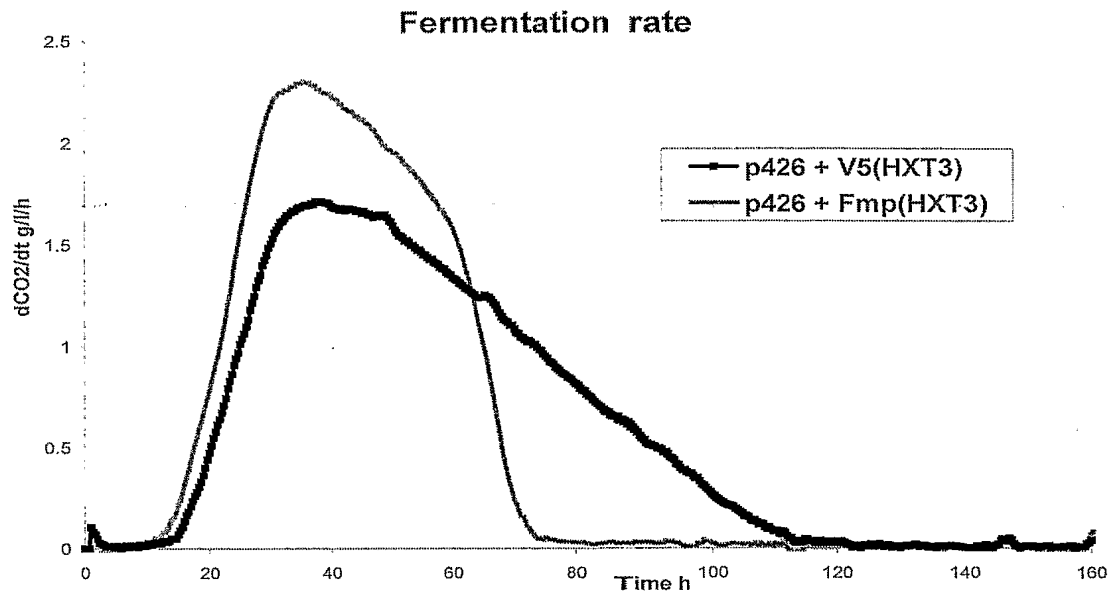
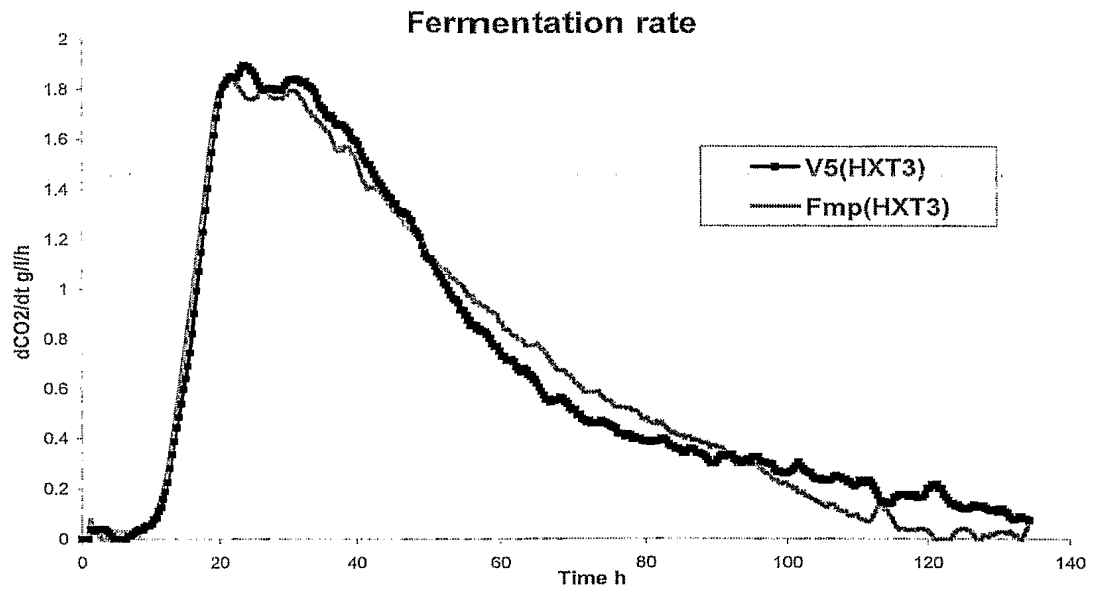Figure 6A : Multicopy overexpression of *HXT3* (V5 or Fmp) on pure Fructose must (200g/l)

Fermentation rate

Figure 6B : Multicopy overexpression of *HXT3* (V5 or Fmp) on pure Glucose must (200g/l)

Fermentation rate

Figure 7A : Single copy expression of *HXT3* (V5 or Fmp) on pure Fructose must (200g/l)

Fermentation rate

Figure 7B : Single copy expression of *HXT3* (V5 or Fmp) on pure Glucose must
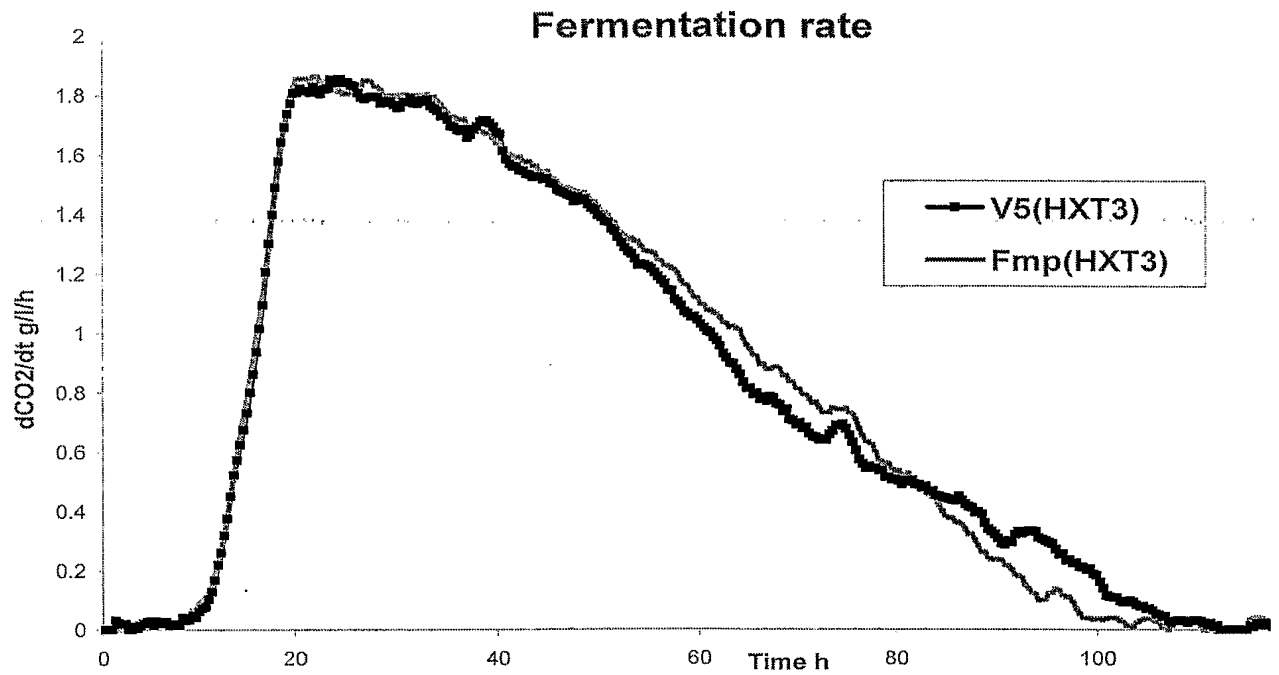(200g/l)

Fermentation rate

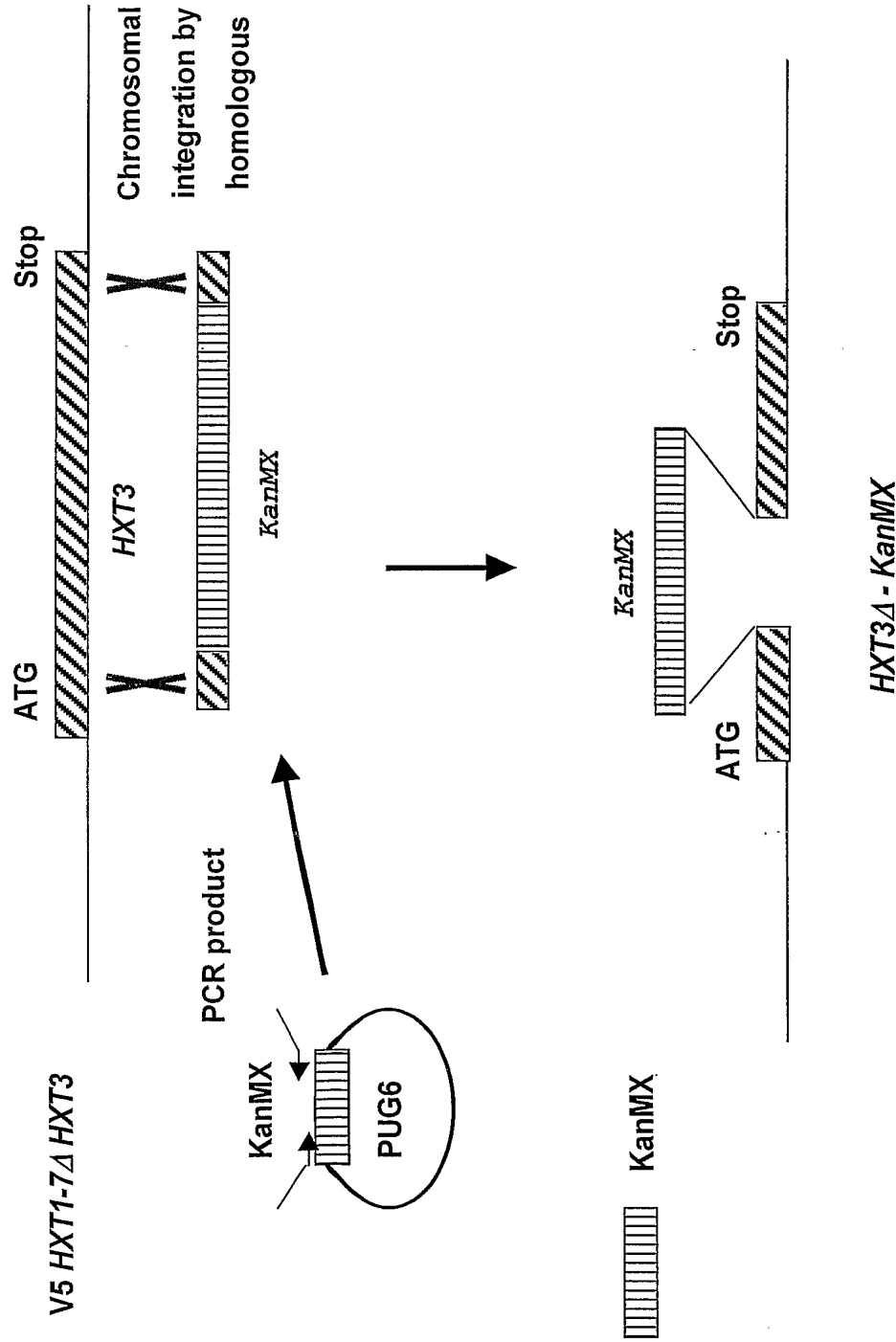Figure 8: Construction of strains that contain a single, inactive, HXT3 gene, general scheme of strains construction

Figure 9: Constructed strains comprising a single, inactive *HXT3* gene
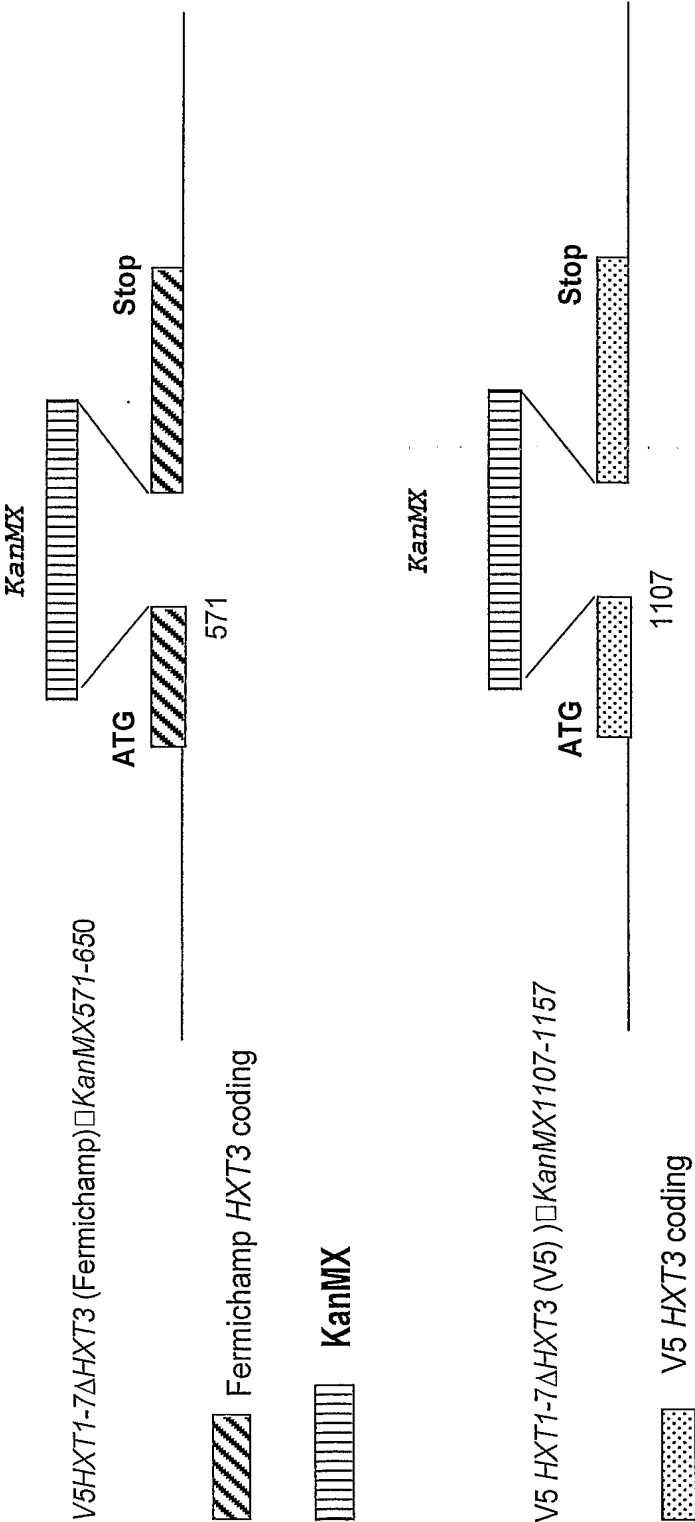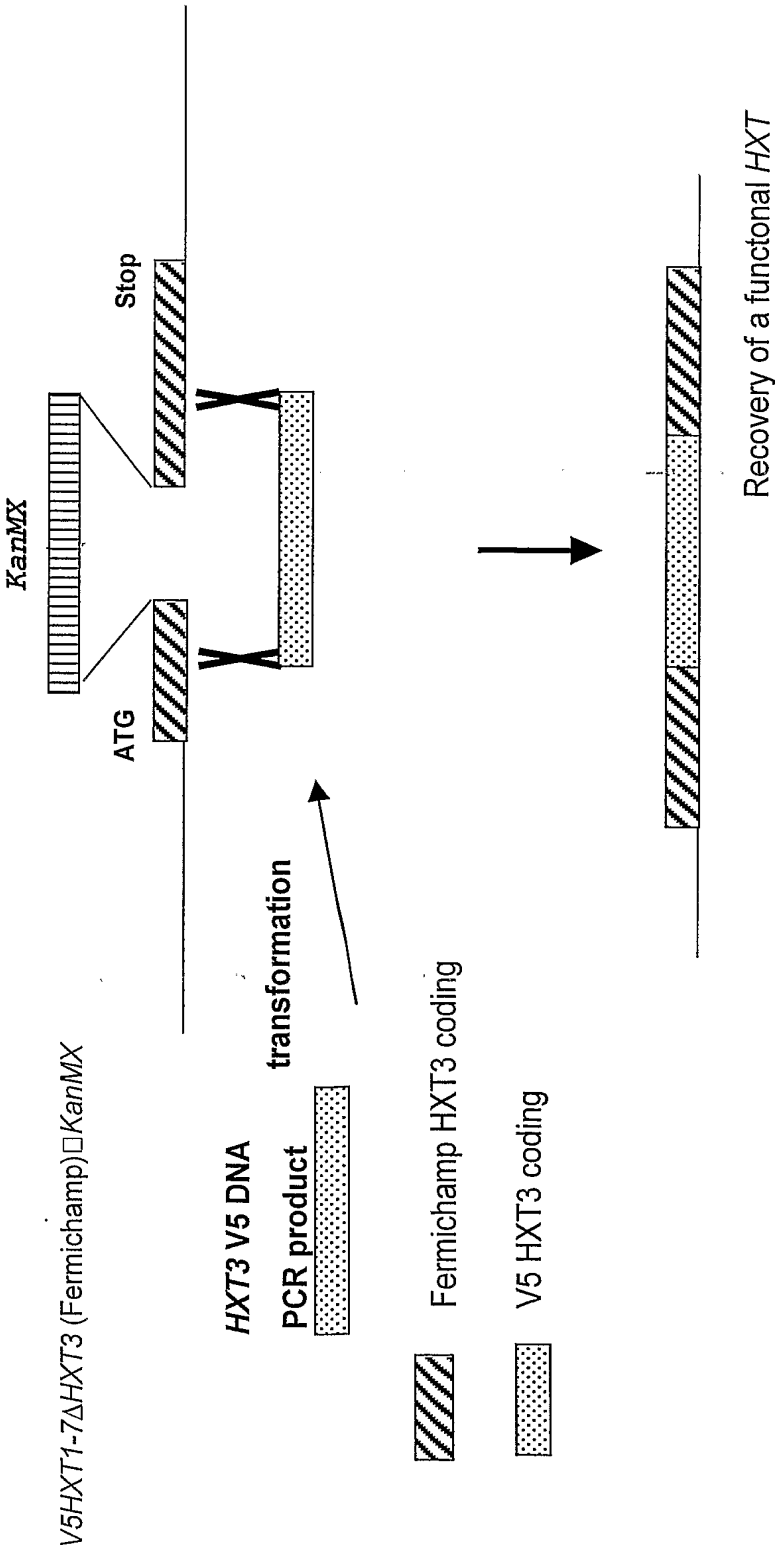
Figure 10: Construction of strains expressing *HXT3* chimera: principle of construction

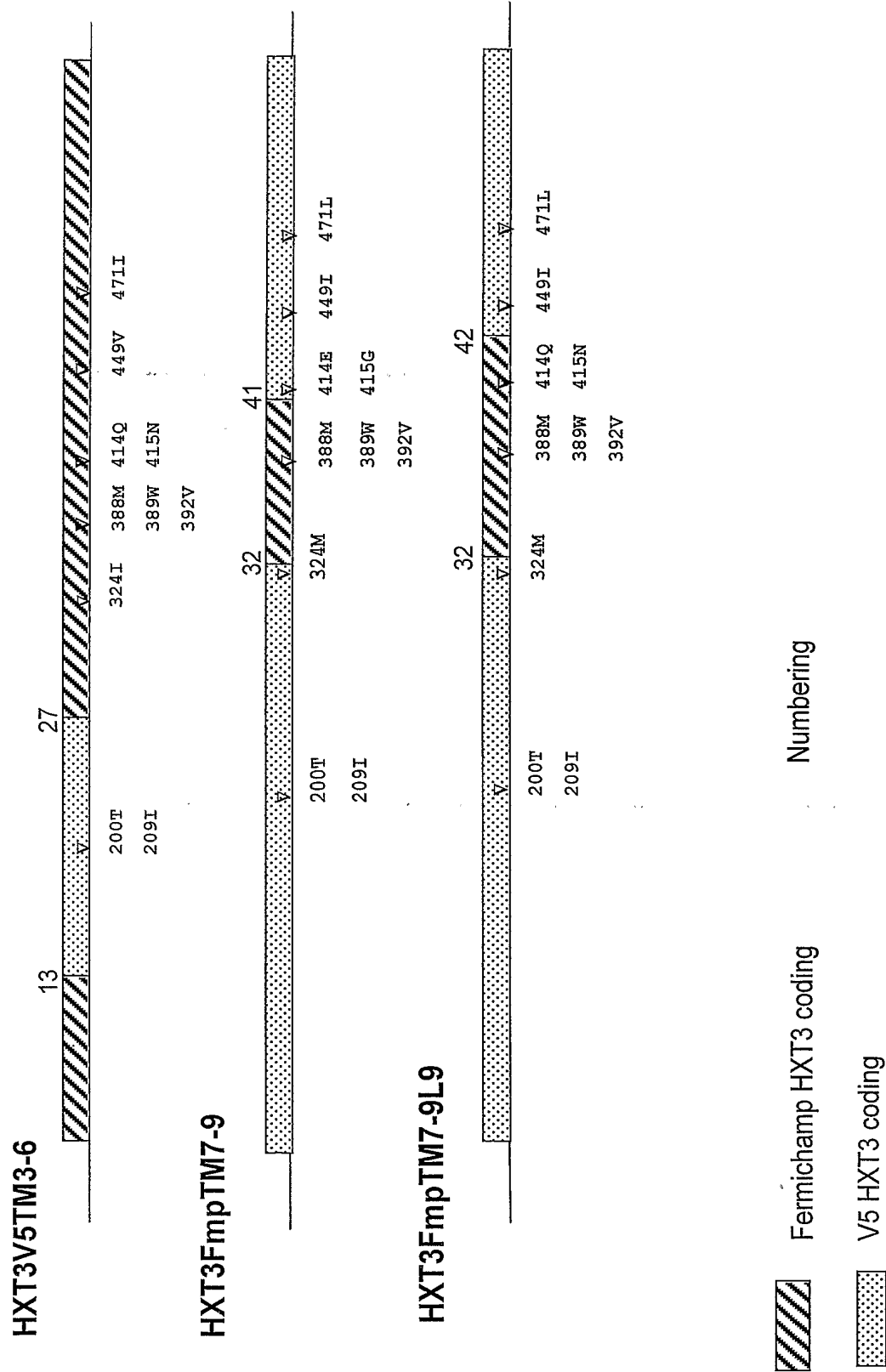Figure 11: Chimeric *HXT3* proteins expressed

Figure 12: Mutated *HXT3* Fermichamp proteins (point mutations)
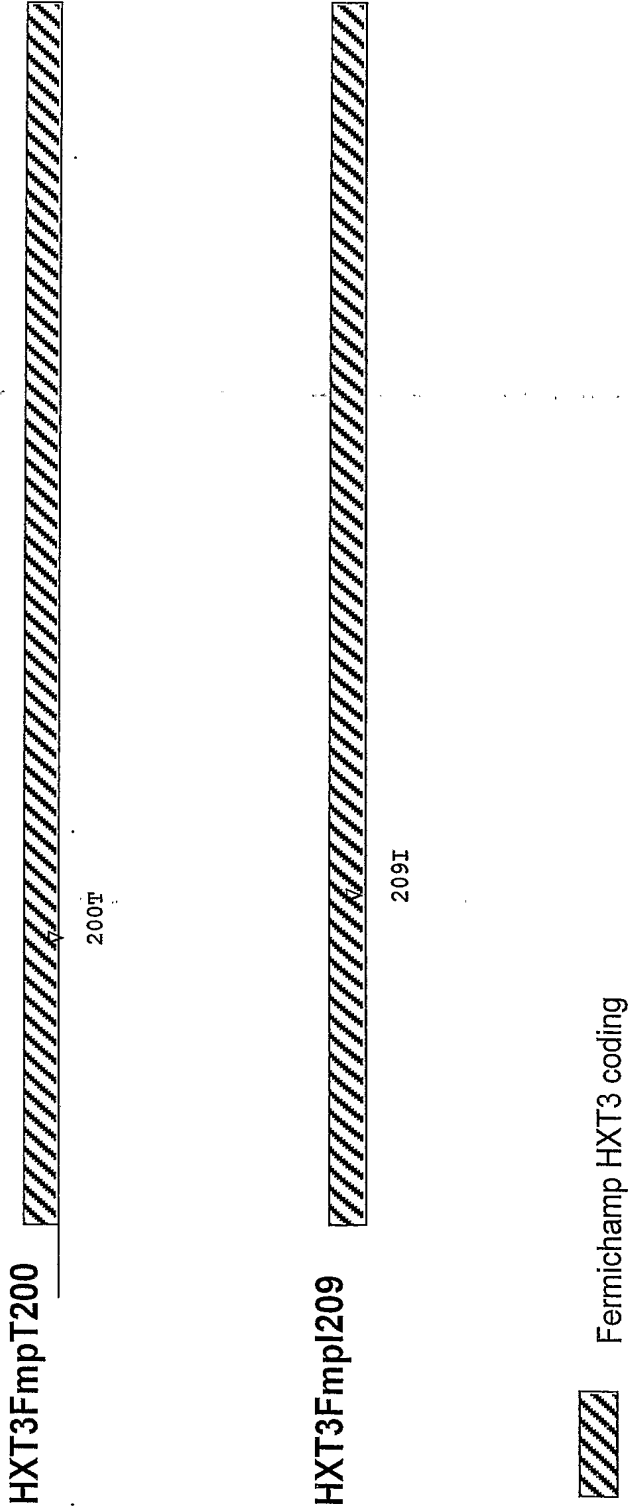
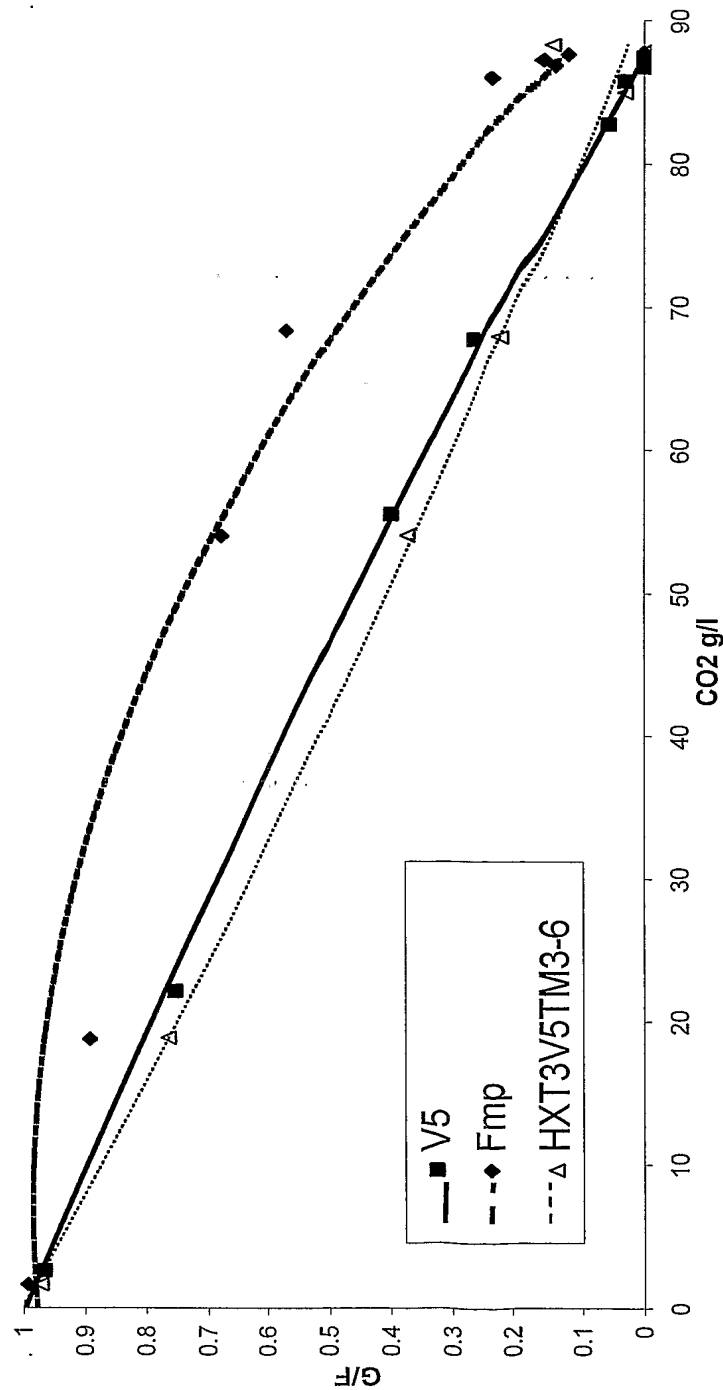Figure 13: Glucose-Fructose ratio evolution during alcoholic fermentation

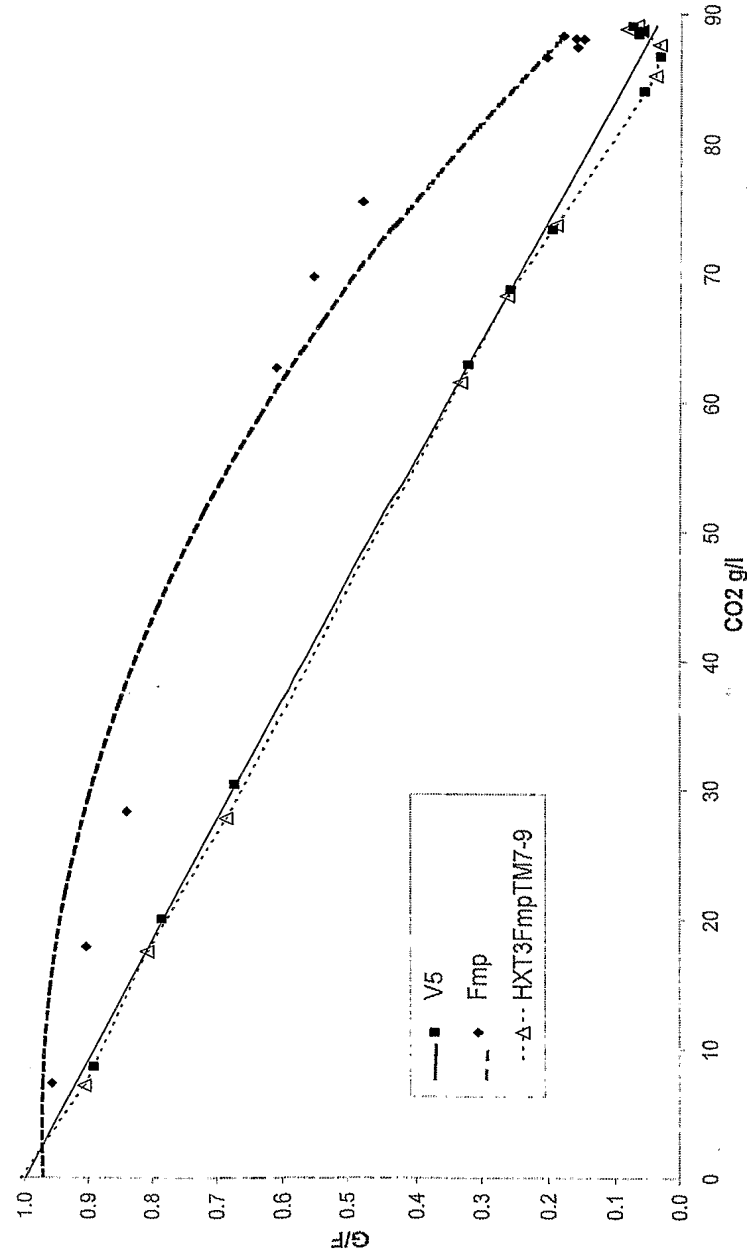Figure 14: Glucose / Fructose ratio evolution during alcoholic fermentation

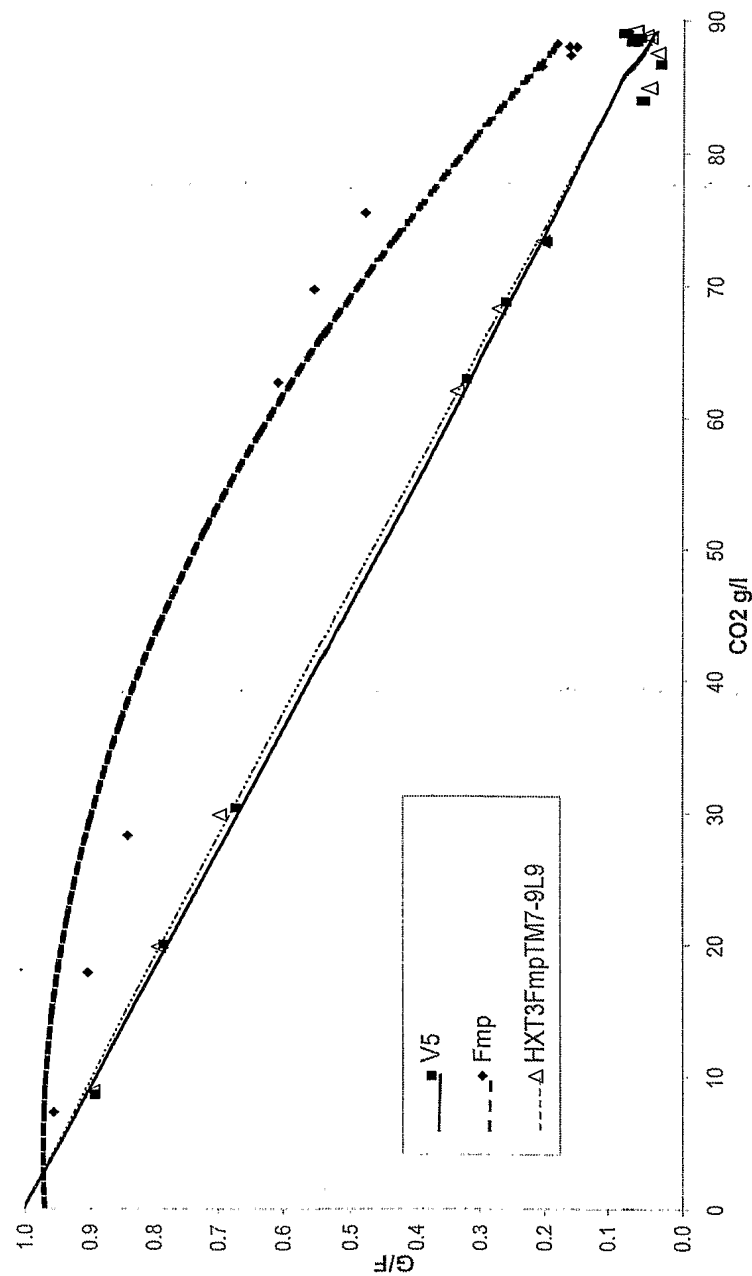Figure 15: Glucose/Fructose ratio evolution during alcoholic fermentation

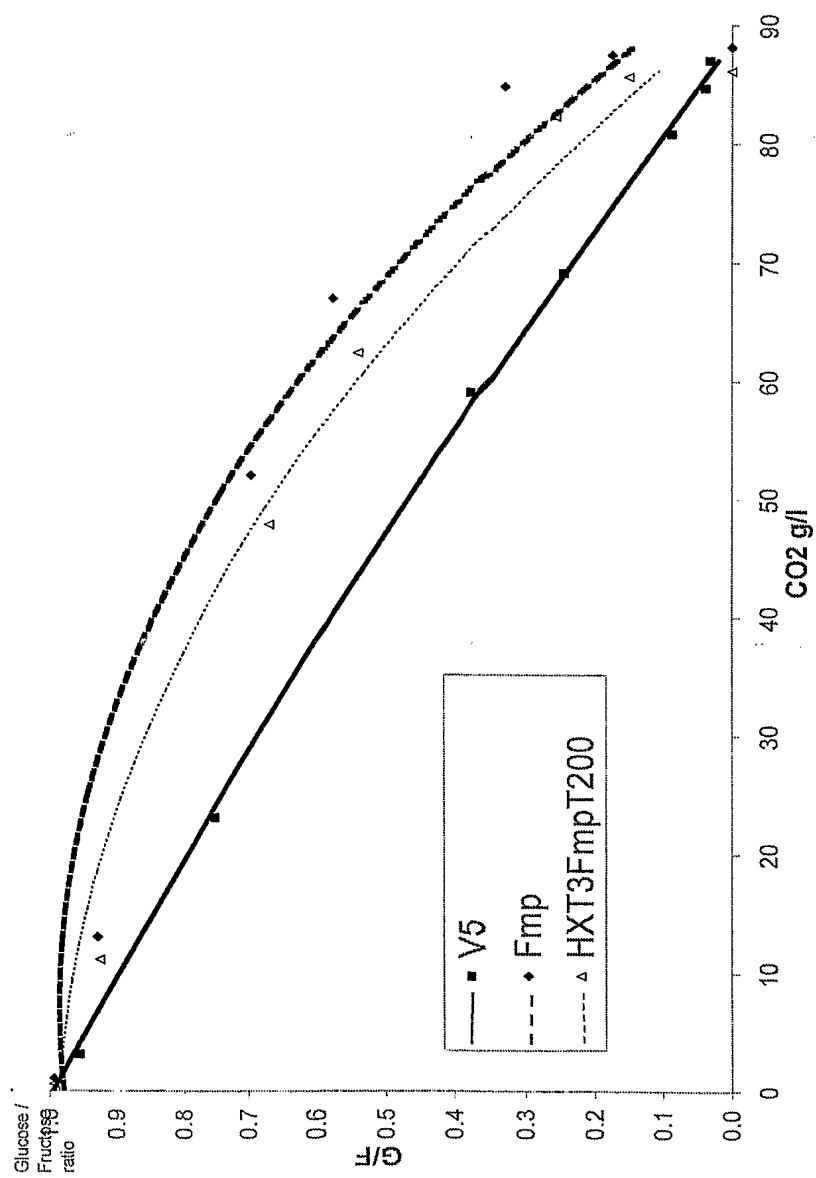Figure 16: Glucose/Fructose ratio evolution during alcoholic fermentation

Figure 17: Glucose/Fructose ratio evolution during alcoholic fermentation